Birds and Microbes:
a story of companionship that has evolved over time

Maisy: How can woodpeckers possibly create nests in solid wood?

Woodpeckers build their homes with the help of fungi. Illustration by Jose Arce Gómez.

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Birds and Microbes

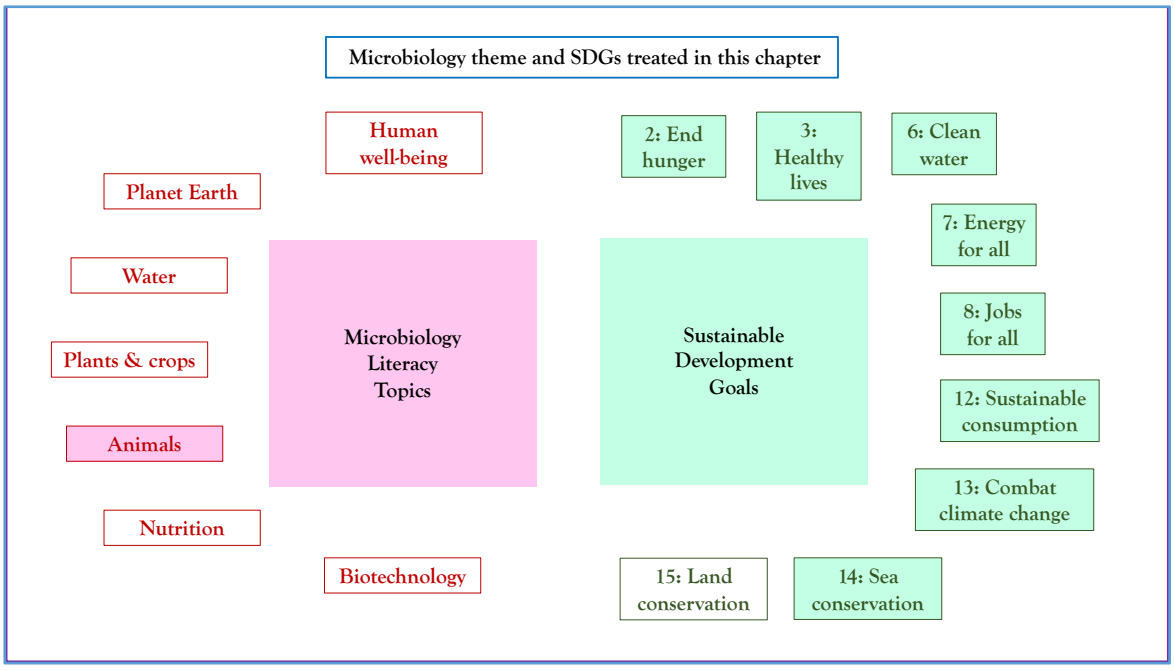
Storyline

Many species of birds live with microbes on their body (plumage, legs, beak and eyes) and within them (stomach and intestine); the vast majority of these are harmless. The interactions allow an exchange of organic matter and molecules between the parties, bringing many mutual benefits. Microbes help birds perform various tasks within an ecosystem, while microbes take advantage of this to spread and feed more easily.

In this lesson, we will find examples of how microbes interact with different species of birds to serve various purposes within an ecosystem. First, we will address the case of the association of microbes with woodpeckers, which help these birds to build their houses. We will also discuss how the woodpeckers and other bird species use their own feathers to line their nests and kill pathogenic microbes. The incredible association between birds and microbes will also be reflected with the discussion of works that show that microbes determine the color of the plumage of certain birds, as well as the importance of the microbes that inhabit the preen glands of birds and emit odors that fulfill the function of improving communication and the search for a mate. Finally, we will discuss about the microbes that inhabit the gastrointestinal tract of birds that play a very important role in their health. All the details of these examples will be seen below!!

The Microbiology and Societal Context

The Microbiology: microbiota; microbial diversity; interactions of microorganisms with birds; symbiosis mechanisms. *Sustainability Issues*: biodiversity loss; climate change; circular economy; energy; food production and safety; environmental pollution; human health; global pandemics.



Microbes and birds: the Microbiology

1. Microbes have been interacting with birds for a long time. Approximately 3.5 billion years ago, microbes appeared as the first and only life forms on our planet Earth. So when birds evolved from dinosaurs 150 million years ago, microbes already knew the secrets of life and the tricks to interact more profitably with these animals. In general, the interactions between microbes and birds have a long evolutionary history, and have played a crucial role in shaping life on our planet.

Many species of birds live with microbes on their bodies (plumage, legs, beak, and eyes) and within them (stomach and intestine), and the vast majority of these interactions are harmless. In general, they allow an exchange of organic matter and molecules between the parties, bringing many mutual benefits. Microorganisms help birds perform various tasks within an ecosystem, while microbes take advantage of birds to spread and feed more easily. Both benefit from this conviviality! Below, we will find examples of how microbes interact with different species of birds to serve various purposes within an ecosystem.

2. Microbes partner with red-crested woodpeckers to help them create their homes. The red-crested woodpecker (*Picoides borealis*) is a cooperative breeder bird that lives in family groups. This endangered bird uses its beak to dig large holes in the bark of trees to form its nest; it is the only bird that chooses live trees instead of dead ones to form these cavities. The work that the woodpecker does requires a lot of energy and time: in general it takes between one year and a decade to complete its home. And the work is even more arduous than this, because each member of the family needs its own nest. However, once they build their houses or nests, these birds can use them for many years.

To reduce the work of building nests in tree stems, red-crested woodpeckers form associations with microbes that have the ability to break down wood. These microbes are fungi that are known as cellulolytic fungi, that is, they have the ability to break down or soften cellulose (cellulose is the main component of tree trunks). These fungi decompose and soften the holes where woodpeckers dig, making it easier for these birds to build their houses; At the same time, these birds help the fungi to disperse and find other sources of nutrients, by transporting the spores on their feathers, legs and beak.

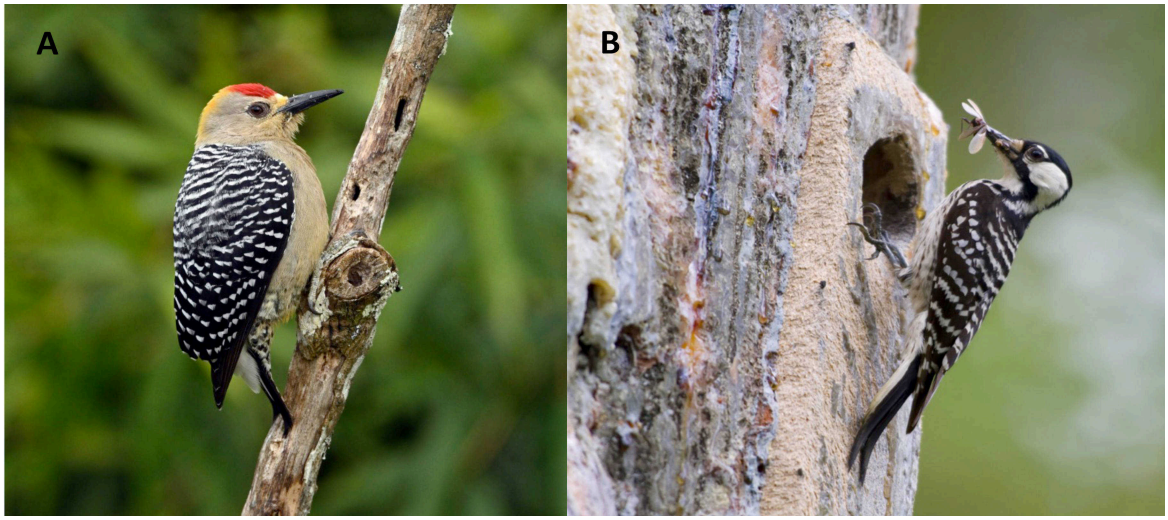
Historically, red-crested woodpeckers were thought to have an association with a single fungus called *Porodaedalea pini*, but recently excavations of these birds have been shown to harbor a diversity of fungi, suggesting that these birds have not one, but many associated fungi. The association between red-crested woodpeckers and fungi is an example of symbiosis. A symbiotic relationship is defined as the joint interaction that two different organisms have, a process of intimate association, the product of an intertwined evolutionary history. They can be of different types, and when in this joint interaction both benefit it is called mutualism.

3. Microbes of red-crested woodpeckers are anti-infective and help reduce disease in the nest. Some pathogenic or disease-causing microbes can affect the health of red-crested woodpeckers when they are in their nests, so these birds have had to develop some defensive strategies. One is association with beneficial microbes with antimicrobial, antiviral and cytotoxic capacities. For example, woodpeckers have been observed to inoculate their nests through their

feces with a fungus called *Acaromyces ingoldii*; this fungus helps them eliminate mites and other ectoparasites, and therefore helps keep their nests clean and free of disease-causing parasites.

A: Hoffmann's woodpecker (*Melanerpes hoffmannii*) is a bird endemic to Central America, resident from southern Honduras to Costa Rica; **B:** Red-crested woodpecker (*Picoides borealis*) lives in the pine forests of the southeastern United States.

Wood-degrading fungi help red-crested woodpeckers build their homes. Red-crested woodpeckers (*Picoides borealis*) live in the pine forests of the southeastern United States. These birds play a very important role as "ecosystem engineers": they form cavities in trees to create their own homes and, when they move, the cavities are used by other species in the ecosystem. The most curious thing about this is that these birds do not work alone in the construction of their houses, but they get help from fungi with wood-degrading capacity. A group of researchers from the Forest Mycology Research Center of the US Forest Service in Wisconsin, experimentally demonstrated a symbiosis between woodpeckers and fungi. They captured woodpeckers in a North Carolina forest and took swab samples from the beaks, legs and wings of captured birds. The team found a wide range of fungal spores, including many that cause wood decay. To see if woodpeckers associate with fungi and transport them to their cavities, the team drilled holes in 60 tree trunks close to other trunks with cavities naturally maintained by woodpeckers. Thirty of these cavities were covered with steel mesh with openings too small for woodpeckers to pass through. After 26 months, only the holes that were accessible to birds had fungal communities living there, and these were similar to those found in the other natural excavations. They concluded that it is possible that the woodpeckers are dispersing the fungi into the cavities to facilitate the work and, furthermore, this demonstrates a complex symbiosis between the two organisms, which may have ecological implications.



Woodpeckers and other bird species have also been observed to use their own feathers to line their nests and kill pathogenic microbes. The feathers act as defenses against pathogenic microbes, thanks to the antimicrobial molecules produced by beneficial microbes that grow naturally in them. The microbes that grow on the feathers of birds are called keratinolytic bacteria, that is, they are capable of degrading keratin, the main component of feathers. Keratinolytic bacteria are microbes that, in addition to producing keratinases, are known for their

ability to secrete broad-spectrum antimicrobial molecules. Woodpeckers and other birds take advantage of this ability and use them in their nests in order to reduce the parasite load and prevent disease.

4. There are microbes associated with birds that determine the color of their plumage. Feathers are the most obvious characteristic of birds and provide various functions, such as providing a high level of insulation, maintaining great flexibility and minimal weight in flight, protection from solar radiation, and waterproofing. But, also importantly, the plumage provides birds with exotic and striking colors. These are key for communication between individuals, camouflage, attraction of mates, defending territory, hiding from predators, and protecting the plumage from solar radiation and environmental abrasion. For example, species of birds that live in the desert, where the sand produces a significant abrasion on the feathers, tend to be black. Melanin serves to absorb ultraviolet radiation, which may explain why birds that live in open areas are also darker in color.

Keratinolytic bacteria usually grow on the plumage of birds and studies have shown that these bacteria can alter the coloration of the feathers, thereby directly affecting secondary sexual characteristics in birds, such as plumage coloration and body condition, both of which are important characteristics in the quest for mates. One study on *Sialis sialis*, commonly called bluebird, showed that female birds had a duller color in their feathers as the number of keratinolytic bacteria increased and, conversely, males tended to be brighter when these bacteria increased. Likewise, the body condition of male birds was positively affected and they had a better biological fitness, that is, a better effect on behavior, reproductive success and survival. Therefore, microbes in birds can be important for fulfilling different roles in the ecosystem, with the growth of harmless microbes in the plumage promoting success in mate attraction and reproduction.

5. The odors emitted by microbes from birds are essential for communication and the search for a mate. Smell is a basic sense vital for the survival of humans and animals. Smell can warn danger, be used to find and enjoy food, and even help to communicate with other individuals and to find a mate. For a long time, it was believed that birds did not have the sense of smell. However, recent studies have determined that birds can not only smell, but even identify each other when they smell the unique aroma created by their microbes that are associated with the oil produced by their preen glands (located at the base of the tail feathers).

Like humans who wear deodorant or perfume, birds groom themselves or become more attractive when they rub their beak over the preen glands and then rub the oil that is produced on their feathers and body. It has always been said that the main function of the oil produced in preen glands is to keep the feathers of birds clean. However, more recently it has been proven that this oil contains microbes that emit unique odors, and that these odors improve communication between birds and the search for a mate. For example, it has been observed that some species of birds show a preference for certain oily odors, and some female birds have even abandoned the father of their offspring in favour of other birds with a 'better' smell.

Microbes that live in bird oil have been found to produce different olfactory molecules. The different combinations of molecules emitted by different birds constitutes custom scents for each bird. This phenomenon is very similar to the situation in humans: we all have microbes on our body that generate odors, for example, armpit odor is generated by microbes that produce a unique odor for each person. The unique odors produced by the microbes associated with birds

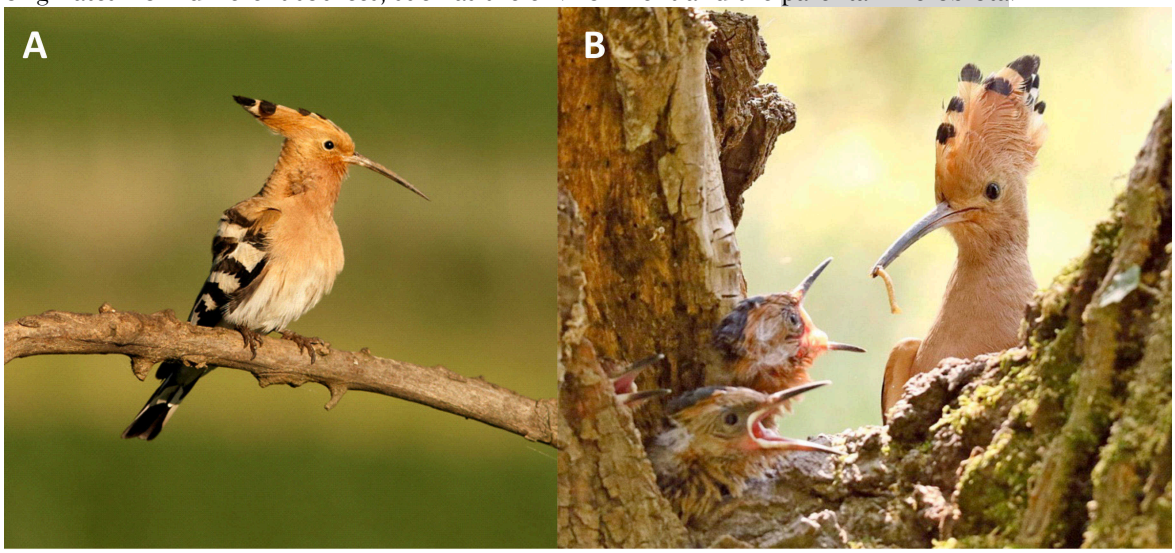
provide crucial information about the stage of the reproductive process – the quality or the hormonal status of a potential mate – directly affecting mating success in birds. An alteration of their microbiota may make potential mates between birds less attractive. This can happen if the environment changes, a bird suffers an infection, or hormonal changes occur.

A: Juncos are the "snowbirds" of the middle latitudes. Over most of the eastern United States, they appear as winter sets in, and then retreat northward each spring; **B:** Bacteria from the "Black-eyed Junco Bird" bird are key to communication and mating in these birds.

Bacteria from the "Black-eyed Junco Bird" are key to communication and mating in these birds. Previous studies have shown that birds use scent to identify other birds and find mates. Recently, scientists at Michigan State University discovered that the scents emitted by the Black-eyed Junco bird (*Junco hyemalis*) are produced by bacteria or microbes that grow in the grooming oil of the preen glands. In addition, an alteration of these odor-producing bacteria could affect the ability of these birds to communicate with each other or find a mate. The scientists did two experimental tests: first they injected antibiotics directly into the preen glands of the birds under study; They observed that when they did this the bacterial communities changed and the most impressive thing was that also the characteristic smell of the Junco bird changed completely. They then isolated the bacteria directly from the grooming oil and measured the odors produced by each of the isolated bacteria. In this second experiment, it was found that all bacteria cultures generated at least one volatile compound (an odor), and that volatile compound was a standard that occurred when grooming oil was measured. The researchers mention that these data experimentally support the hypothesis that there are symbiotic bacteria that live in the grooming oil of the Junco Ojos Negros bird and that they could play a significant role in the production of chemical signals and, therefore, in the behavior of these birds.

A: The Hoopoe bird is a migratory bird that travels from Europe to the south of the Sahara to spend the winter; **B:** It has a gland at the base of its tail from which it secretes a bacteria-laden paste that the mother will spread on her eggs. These microbes help defend the egg and chick against disease and the smelly paste can even ward off predators.

Bacteria that inhabit the preen glands of the "Hoopoe" bird (*Upupa epops*) produce antimicrobial substances against pathogens. The "Hoopoe" is a bird that nests in holes and has a symbiotic association with bacteria that reside in its preen glands. Several studies have shown that these bacteria provide the bird with antimicrobial substances such as bacteriocins, and volatile metabolites that help protect feathers against keratinolytic bacteria, and eggs against pathogens. There is evidence that different species of bacteria inhabit the preen glands of the Hoopoe bird; the microbial community originates from different sources, such as the environment and the parental microbiota.

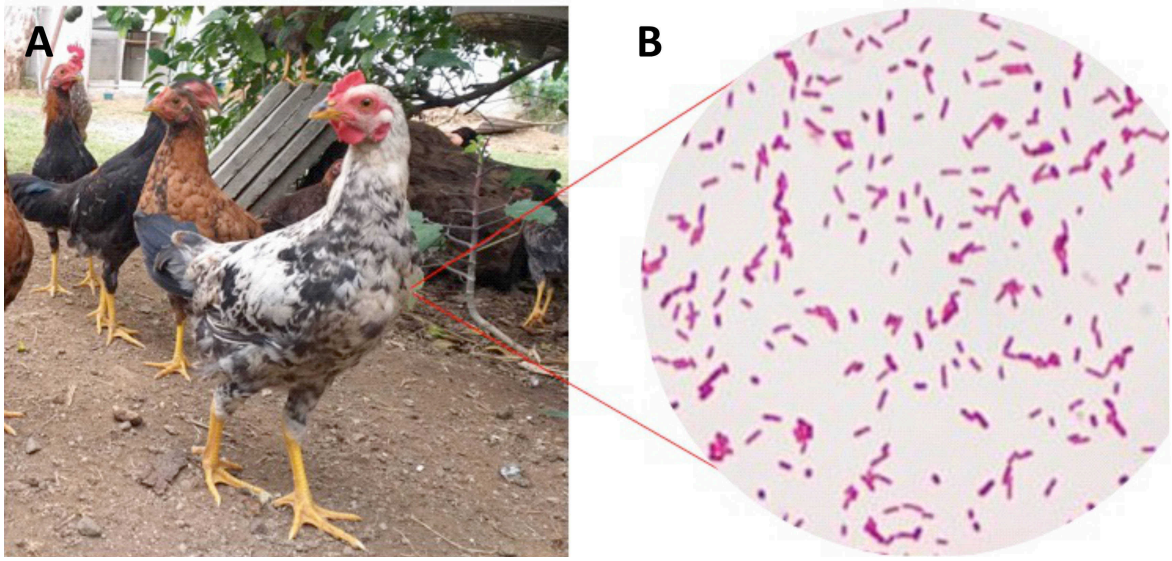


6. Microbes in the gastrointestinal tract play an important role in bird health. The intestine of birds is a complex and dynamic ecosystem made up of various microbes, such as bacteria, fungi, yeasts, and viruses. This ecosystem is known as the microbiota and has a symbiotic relationship with birds. In healthy birds, the microbiota stimulates the immune system, defends the birds against infections, and provides metabolic capacities that allow them to digest and take advantage of components of the food that wouldn´t be metabolized by the digestive system of the birds. Likewise, the microbiota in birds can modulate the expression of different genes, mainly related to the immune system and digestive physiology. In itself, the microbiota has a great capacity for adaptation and quite remarkable resilience, but it can be altered by various factors and it takes a long time to return to the original state of equilibrium.

Studies on the influence of the microbiota on poultry health and production have increased in recent years and new approaches to analyze and interpret the functioning of the microbiota in poultry has made it possible to follow the evolution of these microorganisms in response to changes in diet, stress, treatment with antimicrobials, probiotics, etc., opening a new field of work that contributes significantly to improving the health and productivity of poultry production.

A: Domestic and farm poultry, such as chickens, often suffer infections of multiple *Salmonella* bacteria**;**
B: *Salmonella* are pathogenic bacteria that cause severe diarrhea in humans.

Alert! Not all bird-related bacteria are good for the birds and us. Even though some bacteria that live with birds provide benefits, there are also some that may be dangerous for them and us. This is particularly true of domestic birds (Aviculture). For instance, chicken-derived products are strongly associated with the presence of pathogenic bacteria such as *Salmonella*. The best way to avoid getting sick from these bacteria is to sanitize eggs and other chicken-related products before consuming them



Relevance for Sustainable Development Goals and Grand Challenges

- **Goal 2. End hunger.** Even though most of the bird:microbe associations treated here involve wildlife, the association between microbes and aviculture birds is significant in order to in increase the food production. Understanding that the microbiota of these birds impact directly its development will help in the development of novel alternative for aviculture that generate less pollution.
- **Goal 3. Ensure healthy lives and promote well-being.** By understanding that microbes are a significant part of birds' body; we also understand the care that must be taken when interacting with wildlife which may harbour microbes that cause zoonotic diseases.
- **Goal 6. Clean water and sanitation.** Understanding the interaction of birds with the microbiota can help us reduce the use of antibiotics and other chemical products in poultry farms, which lead to contamination of waters and soils.
- **Goal 7. Affordable and clean energy**: Particularly woodpeckers are associated with multiple cellulolytic fungi. Cellulose is an abundant renewable source of carbon that, once degraded, for example by cellulolytic fungi, into simple sugars, can be used to

produce other chemicals such as biodiesel, which has great potential as a clean energy source.

- **Goal 8. Jobs for all.** Bird watching is a human activity that has expanded over the entire world and has gained economic importance in some countries. Understanding the functional importance of bird microbiota may help to improve conservation practices that enable proliferation of endangered species and at the same time increase the touristic attraction of a region.
- **Goal 12. Eliminate pollution**. Aviculture wastes are usually not well managed and do not have a standardized way of treatment. On the other hand, cellulolytic fungi are important for recycling multiple agriculture wastes.
- **Goal 14. Life below water.** Use of fewer chemicals in lower quantities in aviculture will reduce contamination of rivers, lakes and oceans. Additionally, understanding the importance of birds in their habitats will help to make people aware of the importance of reducing the use of polluting chemicals in their daily life.

Potential Implications for Decisions

1. ***Individual***

a. How close should we be to wild birds? We need to understand that birds carry their own microbiota that can help them survive in their environment and we must avoid perturbing it. And, on the other hand, there are associated risks to humans due to zoonotic diseases.

b. Encourage individual sustainable consumption patterns and lifestyles to reduce environmental footprints, for example by making informed decisions concerning the use of avicultural products based on an environmental friendly principles, and avoiding non-clinical use of antibiotics that increase health problems and antimicrobial resistance.

2. ***Community policies***

a. How to protect birds? Increase awareness about the role of birds and their microbiota to promote creation of community policies that protect locally-endangered species, and thereby across the globe. Awareness also creates consciousness regarding the hunting and capture of exotic species.

b. Increase awareness that eggs and other bird-derived products carry multiple bacteria that may be pathogenic to humans to improve community hygiene habits and reduce the rate of infection of bird-derived pathogens, such as *Salmonella*

3. ***National policies***

a. Understanding the importance of birds and their microbiota in natural environments helps raise awareness about illegal hunting and encourage the generation of new measures to eliminate this practice that affect multiple endangered species.

Pupil Participation

1. ***Class discussion of the issues associated with bird microbiota***
    a. Do microbes live in or on birds? How do these microbes influence their health?

b. Discuss how woodpeckers interact with fungi in order to build their homes? What do the fungi get in exchange?

c. How do microbes affect the way birds communicate with one another? Where do the birds obtain their microbes?

2. ***Pupil stakeholder awareness***

a. Should we be careful when we consume bird-associated products? Can you name some bacteria commonly found in such products?

b.

3. ***Exercises***

a. Would you say that the woodpecker:fungi interaction is a symbiosis or a mutualistic interaction? Explain the difference.

b. Investigate woodpeckers on the web. How long do they need to build their nests? Is this enough time for fungi to degrade the wood in the tree?

c. Go outside: can you find any bird? According to what you see can you try to guess what this bird feeds on and how this food may influence the microbiota they house?

d. Investigate another animal whose colour may be determined by its microbiota. What do you learn?

The Evidence Base, Further Reading and Teaching Aids

Video: The Stinky Shield of the Hoopoe Bird | Microbe Minute
/https://www.youtube.com/watch?v=8GPEs5ldBmc

Video: Expedition 1_Microbes in Bird Bath
/https://www.youtube.com/watch?v=KDWdxbcdmgY

Video: Expedition 52_Microbes in Bird Bath S2 /
https://www.youtube.com/watch?v=uoPIc83L7_g

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Glossary

Fungi: Eukaryotic spore-forming microorganisms that feeds in organic matter ie. molds, yeast and mushrooms
Symbiotic relationship: A relationship between two organisms of different species in which both of them need the other one in order to survive.
Mutualism: A relationship between two organisms of different species in which both of them benefit from their association but is not strictly required for their survival
Cellulolytic fungi: Fungi that degrade cellulose using extracellular enzymes that break their carbon bounds to produce simpler sugar such as glucose
Cellulose: Polymer produced by plants, it consists in a polysaccharide base on linear chain of glucose molecules.
Spores: a unicellular reproductive body which is usually dormant and present an increase resistance to environmental stresses.
Pathogenic: biological agent that causes a disease to its host
Antimicrobial: Compounds that are able to inhibit the growth of microorganism due to their chemical nature
Antiviral: A chemical compound that is able to inhibit the replication of virus.
Ectoparasites: Parasites that infect the Surface of its host, such as skin. Fleas and thicks are classified as ectoparasites.
Keratinolytic bacteria: Keratin is the main protein of the hair and feathers. keratinolytic bacteria are the microbes that are able to feed on the keratin
Parasite: A organism that live in/on another specie and nurture by affecting negatively its host.
Camouflage: The ability to hide or disguise of an organism in a determined environment.
Preen glands: A gland at the base of the tail of birds that produce preening oil.
Bacteriocins: A bacterial protein that inhibit the growth of usually phylogenetically close related bacteria.

Volatile metabolites: Chemical compounds that are characterized for their ability to vaporize at relative low temperatures.
Fluorescence microscope: A microscopy technique where fluorescent substances are examined using a microscope.
Hoescht dye: Fluorescent dye that stains DNA in bacteria and eukaryotic cell
Microbiota: the set of all the microorganisms that inhabit a specific habitat.
Immune system: A set of organs and cells that are in charge of protecting the body from pathogenic infections.
Probiotics: Bacteria that benefit the gastroinstestinal system of their host by the secretion of multiple compounds